(CONVENTION. By one or more persons and/or a Cor

m 4

2

COMMONWEALTH OF

Patents Act 1952-1

CONVENTION APPLICATION FOR A PATENT

83148/75

(1) Here insert (in full) Name or Names of Applicant or Applicants, followed by Address (es).

We, HOECHST AKTIENGESELLSCHAFT,
of 45 Bruningstrasse,
Frankfurt/Main,
Federal Republic of Germany

hereby apply for the grant of a Patent for an invention entitled: (2)
PEPTIDECYCLOPROPYLAMIDES HAVING LH-RH/FSH-ACTIVITY

(2) Here insert Title of invention

which is described in the accompanying complete specification. This application is a Convention application and is based on the application numbered (3)
P 24 38 352.6
for a patent or similar protection made in (4) Germany
9th August, 1974

(3) Here insert number(s) of prior application(s)

(4) Here insert Name of basic Country or Countries, and basic date or dates

Our address for service is Messrs. Edwd. Waters & Sons, Patent Attorneys, 50 Queen Street, Melbourne, Victoria, Australia.

DATED this 6th day of August 19 75

(5) Signature (s) of Applicant (s)

Seal of Company and Signature of its Officers as prescribed by its Articles of Association

HOECHST AKTIENGESELLSCHAFT
by J. A. Barnes

To:

THE COMMISSIONER OF PATENTS.

Edwd. Waters & Sons, Melbourne.

COMMONWEALTH OF AUSTRALIA Form 7

Patents Act 1952

DECLARATION IN SUPPORT OF A CONVENTION APPLICATION
UNDER PART XVI. FOR A PATENT.

8374/75

In support of the Convention application made under Part XVI. of the Patents Act 1952 by HOECHST AKTIENGESELLSCHAFT

of 45, Brüningstrasse, Frankfurt/Main, Federal Republic of Germany

for a patent for an invention entitled:
PEPTIDECYCLOPROPYLAMIDES HAVING LH-RH/FSH-ACTIVITY

We, Albert Schüler, Horst Grove of 17,Fuchshohl,Neuenhain/Taunus, 15,Weinbergstrasse,Wiesbaden; Federal Republic of Germany

do solemnly and sincerely declare as follows:

1. We are authorized by HOECHST AKTIENGESELLSCHAFT

the applicant for the patent to make this declaration on its behalf.

2. The basic application as defined by Section 141 of the Act was made at München in the Federal Republic of Germany under No. P 24 38 352.6

on the 9th day of August 19 74 by HOECHST AKTIENGESELLSCHAFT

3. 1) Wolfgang König, 25, Eppsteiner Strasse, Langenhain/Taunus
2) Rolf Geiger, 33, Heinrich-Bleicher-Strasse, Frankfurt/Main
3) Jürgen Kurt Sandow, 43, Strrbbergstrasse, Frankfurt/Main
1) - 3) Federal Republic of Germany

~~is~~/are the actual inventor(s) of the invention and the facts upon which HOECHST AKTIENGESELLSCHAFT

is entitled to make the application are as follows:

The said HOECHST AKTIENGESELLSCHAFT

is the assignee of the said Wolfgang König, Rolf Geiger and Jürgen Kurt Sandow

4. The basic application referred to in paragraph 2 of this Declaration was the first application made in a Convention country in respect of the invention the subject of the application.

DECLARED at Frankfurt/Main, Federal Republic of Germany

this 28th day of July 1975.

To the Commissioner of Patents

PAT.

Hoechst Aktiengesellschaft

Prokurist (ppa.Schüler) Handlungsbevollmächtigter (i.V. Grove)

PAT 510
8/86

Form 10

COMMONWEALTH OF AUSTRALIA

PATENTS ACT 1952-69

COMPLETE SPECIFICATION

(ORIGINAL)

Application Number: 52172/75

Lodged: 7/8/75

Class: Int Cl C07C 103/52 461K 31/195

Int. Class: 79.65-1 87.16

Complete Specification Lodged: 7/8/75

Accepted: Lapsed Sec 52 B(3)

Published: 10/2/77

Priority: (33) (31) (32) 9/8/74 P24 38 352.6

Related Art:

Name of Applicant: HOECHST AKTIENGESELLSCHAFT,

Address of Applicant: 45 Bruningstrasse, Frankfurt/Main, Federal Republic of Germany

Actual Inventor: WOLFGANG KONIG, ROLF GEIGER and JURGEN KURT SANDOW.

Address for Service: EDWD. WATERS & SONS, 50 QUEEN ~~VICTORIA~~ STREET, MELBOURNE, AUSTRALIA. 3000.

Complete Specification for the invention entitled:

PEPTIDECYCLOPROPYLAMIDES HAVING LH-RH/FSH-ACTIVITY

The following statement is a full description of this invention, including the best method of performing it known to :- us

C. J. 5/1/78

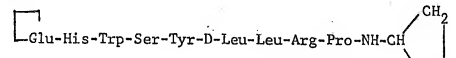
The present invention relates to peptide cyclopropyl amides having LH-RH/FSH activity.

Analogues of the hypothalamus hormone LH-RH of the formula

$$\text{Glu-His-Trp-Ser-Tyr-Gly-Leu-Arg-Pro-Gly-NH}_2$$
$$1 \quad 2 \quad 3 \quad 4 \quad 5 \quad 6 \quad 7 \quad 8 \quad 9 \quad 10$$

in which the glycin amide is replaced by ethyl, propyl or isopropyl amide, have increased biological activity according to J.Med. Chem.16 (1973), page 1144. In a similar manner, the activity of the hormone is increased when glycin is replaced in 6-position by D-alanin. When Gly^6 is replaced by D-valin, D-leucin or D-prolin, the activity decreases as compared with the natural hormone (cf. Abstr. of the Endocrine Society, 55th Ann. Meeting, 1973, page A 145). According to Biochem. Biophys. Res. Commun. 57 (1974), page 335, the biological activity is further increased, when the replacement of Gly^6 by D-ala is combined with the replacement of $Glycinamide^{10}$ by the above-mentioned amides.

This invention relates to peptides of the general formula I



(I)

in which, optionally, Tyr may be replaced by Phe and Leu^7 by $Ser(Bu^t)$, $Asp(OBu^t)$, $Glu(OBu^t)$, $Cys(Bu^t)$, Lys(Boc) or Orn(Boc).

In the ovulation test on rats, peptides of the general formula I in which no amino acid elements are exchanged, surprisingly show an extremely strong biological activity. This test is the best criterion for the practical utility of such a peptide, because it displays the intergral of the time-activity-curve. The time during which the LH-release occurs shows that

this strong activity is not only due to the intensity but also, and above all, to the unexpectedly prolonged duration of activity. About the same integral activity is obtained when additionally Tyr^5 is replaced by Phe and/or Leu^7 by $Ser(Bu^t)$, $Asp(Bu^t)$, $Glu(OBu^t)$, $Cys(Bu^t)$, Lys(Boc) or Orn(Boc). Above all the exchange in 7-position causes a significant additional prolongation of the LH- and FSH-releasing activity of these compounds.

Moreover, these peptides of the formula I also have peroral activity. This surprising effect, so far undetected in any peptide of this chain-length, opens new possibilities of therapy and makes the compounds of the invention superior to all LH-RH-analogues that have been prepared until now. Compounds of the formula I in which Leu^7 is replaced by Lys(Boc) or Orn(Boc), have only parenteral activity, because in these two amino acid derivatives the Boc-group is not sufficiently stable against gastric acid. However, analogues having amino acid derivatives in 7-position which contain tert.butyl ether and -ester groups, have oral activity, although these groups are capable of being split off, too, in an acid medium.

This invention further relates to compositions for peroral, intranasal, intramuscular, subcutaneous or intravenous adminstration of the peptide of the formula II and of the above-mentioned analogues as well as to processes for manufacturing them.

Further object of this invention is a process for the preparation of the above peptides, which comprises preparing them

a) by fragment condensation of peptide fragments usual in peptide chemistry according to the condensation scheme

1 - 3 + 4 - 10 or 1 - 2 + 3 - 10 or

b) by stepwise synthesis,

in which case other functional groups may be blocked intermediately by protective groups capable of being split off by hydrogenation or capable of being split off in an alkaline or slightly acid medium.

As compounds in which the exchange of amino acids in 5- and 7-position occurs according to the invention, there are considered, for example, the following ones:

1	2	3	4	5	6	7	8	9
Glu-	His-	Trp-	Ser-	Tyr-	D-Leu -	Ser(Bu^t)	-Arg-	Pro-NH-CH(CH_2-CH_2)
				Phe		Ser(Bu^t)		
						Cys(Bu^t)		
						Asp(OBu^t)		
						Glu(OBu^t)		
				Phe		Asp(OBu^t)		
				Phe		Glu(OBu^t)		
				Phe		Cys(Bu^t)		

In the synthesis of the compounds of the formula II in which Leu is not replaced, the use of protective groups is not critical. As intermediary α-amino protective groups are considered, for example, the benzyloxycarbonyl or the tert.-butyloxycarbonyl radical. The guanido function of the Arginin may remain unprotected or it may be blocked by a nitro group which is split off in the following hydrogenation. The same applies for the benzyloxycarbonyl group. The tosyl radical can also be used as guanido protective group. It can be removed, optimally together with further protective groups capable of being split off by acids, after the synthesis being finished or at an earlier stage of the synthesis by HF/anisol. The nitro group is also split off by HF/anisol and can therefore be used, for example in combination with

the ter.-butyloxycarbonyl radical, also as α-amino protective group during the whole synthesis for the protection of the guanido function.

When Leu^7 is replaced by an amino acid derivative having a tert.-butyl radical unstable towards acids, the nitro- or the tosyl group must be split off by HF/anisole already at the dipeptide stage. The same is true for an intermdiary protection of the amide group.

In this case, suitable intermediary α-amino protective groups are those which can be split off by hydrogenation, for example the benzyloxycarbonyl radical or groups capable of being split off by weak acids, for example, the 2-(p-diphenyl)-isopropyloxy-carbonyl- or the 2-(3,5-dimethoxyphenyl)-isopropyloxycarbonyl radical.

In the synthesis of sulfur-containing compounds, benzyloxycarbonyl protective groups must be split off by catalytical hydrogenation in known manner by adding an amine, for example, triethyl amine, N-ethyl morpholine or cyclchexyl amine, because the hydrogenolytical removal of the protective group in a neutral or an acid medium is not advantageous in this case.

In fragment coupling according to a), the azide coupling performed without racemization used or the DCC/1-hydroxybenzotriazole- or the DCC/3-hydroxy-4-oxo-3.4.-dihydro-1.2.3-benzotriazine method is preferably used. Activated esters of fragments can also be used.

For the stepwise condensation of amino acids according to b) especially suitable are activated esters of benzyloxycarbonylamino acids, for example N-hydroxysuccinimide esters or 2.4.5-trichlorophenyl esters and 4-nitrophenyl esters. The aminolysis

of the two latter active esters can very well be catalized by N-hydroxy compounds of about the acidity of the acetic acid, for example 1-hydroxybenzotriazole.

In the ovulation test and in the ascorbic acid depletion test, the compounds of the invention show a stronger and prolonged activity as compared with the 6-Gly analogues, but also with the 6-D-Ala-analogues.

Due to the oral activity, these medicaments have a large field of application than the LH-RH which could only be administered, so far, via the parenteral or the nasal route. They are novel medicaments which cause in the case of insufficiency of hypothalamus and hypophysis the release of the luteinizing and the follicle stimulating hormone from the anterior lobe of the hypophysis and are, therefore, used for the treatment of female and male sterility, as far as this sterility has a hypothalamic -hypophyseal origin. The compounds of the invention can also be applied for the determination of the ovulation time for women. Shortly before the expected ovulation time an ovulation can be released for certain by the administration of the new medicaments. This is an important fact for family planning according to the Knaus-Ogino methods as well as for artificial insemination.

The compounds of the invention can be administered via the intravenous, the intramuscular or the subcutaneous route, via the intranasal route in the form of nose drops or nose sprays and, with the restriction mentioned above, also via the peroral route. For the different adminstration forms the following doses are preferably used:

20	-	1,000 ng/kg	intravenous
20	-	2,000 ng/kg	subcutaneous

20	-	10,000 ng/kg	intramuscular
100	-	50,000 ng/kg	intranasal
10,000	-	200,000 ng/kg	peroral

The following Examples illustrate the preparation of the compounds of the invention. They could all be synthesized according to one of the methods of the invention.

The peptides have been examined for uniformity according to thin-layer chromatography and characterized by optical rotation, amino acid analysis and OCH_3-determination. The values calculated and found for the peptides are very similar.

Abbreviations:

DCC	=	dicyclohexyl carbodiimide
HOBt	=	1-hydroxybenzotriazole
OOBt	=	3-hydroxy-4-oxo-3.4-dihydro-1.2.3-benzotriazine ester

E X A M P L E 1

Glu-His-Trp-Ser-Tyr-D-Leu-Leu-Arg-Pro-cyclopropyl amide

a) Z-Pro-cyclopropyl amide

To a solution of 25 g (0.1 mole) of Z-prolin, 13.5 g (0.1 mole) of HOBt and 5.71 g (0.1 mole) of cyclopropyl amide in 200 ml of absolute tetrahydrofurane were added at 0°C 22 g of DCC, dissolved in 50 ml of cold absolute tetrahydrofurane. The solution was allowed to stand for an hour at 0°C and for 3 hours at room temperature, the precipitate was suction-filtered and the filtrate was concentrated. The oily residue was dissolved in ethyl acetate and successively shaken with saturated $NaHCO_3$-solution, 2 N HCl, $NaHCO_3$-solution and water, dried over Na_2SO_4 and concentrated. The residue was triturated with petroleum ether and suction-filtered. It was purified by recrystallization from ethyl acetate/

petroleum ether, Yield: 23 g (= 80 %).

Melting point: 120 - 123°C, $[\alpha]_D^{24}$ = -45.5° (c=1, in methanol)

b) H-Pro-cyclopropyl amide · HCl

19 g of Z-Pro-cyclopropyl amide were dissolved in 200 ml of methanol. A spatula tip of Pd/$BaSO_4$-catalyst was added and the solution was hydrogenated by allowing hydrogen to pass through the solution while stirring. The pH of the solution was maintained at 4.5 with the aid of an autotitrator by adding 1 N methanolic hydrochloric acid. The catalyst was suction-filtered after hydrogenation being finished and the filtrate was concentrated. The residue was triturated with ether and suction-filtered. Yield: 11 g (3 38 %)

Melting point: 169 - 173°C.

c) Z-Arg(Z_2)-Pro-cyclopropyl amide

To a solution of 28.85 g (50 moles) of Z-Arg(Z_2)-OH, 9.5 g (50 mmoles) of H-Pro-cyclopropyl amide. HCl and 6.75 g (50 mmoles) of HOBt in 100 ml of methylene chloride and 25 ml of dimethylformamide were added 6.5 ml of N-ethyl morpholine and at 0°C a solution of 11 g of DCC in a small amount of methylene chloride. The solution was allowed to stand for an hour at 0°C and overnight at room temperature. The precipitate was suction-filtered and the filtrate was concentrated. The residue was taken up in ethyl acetate and washed with water, $NaHCO_3$-solution, 1 N HCl and $NaHCO_3$-solution, dried with Na_2SO_4 and concentrated. The residue was recrystallized from ethyl acetate/petroleum ether. The crude substance (26.3 g) was purified chromatographically on a 250 g silica gel column in methylene chloride/acetone in the ratio 9:1 and 8:2. Yield: 22.2 g (62 %).

Melting point: 171°C, $[\alpha]_D^{21}$ = -33.0° (c=1, in methanol)

d) H-Arg-Pro-cyclopropyl amide · 2 HCl

22 g (30.9 mmoles) of Z-Arg(Z_2)-Pro-cyclopropyl amide were hydrogenated catalytically in methanol according to b).

The residue was dried under highly reduced pressure. 11 g (89 %) of an amorphous substance were obtained.

e) Z-D-Leu-Leu-Arg-Pro-cyclopropyl amide

To a solution of 3.83 g (10 mmoles) of H-Arg-Pro-cyclopropyl-amide · 2 HCl in 20 ml of dimethyl formamide were added at 0°C 2.6 ml of N-ethyl morpholine and 2.75 g of Z-Leu-ONSu. The solution was allowed to stand overnight at room temperature, it was concentrated and the residue was trifurated once with ethyl acetate and once with ether. The solvents were decanted and the oil was dried under highly reduced pressure.

The residue was dissolved in methanol and hydrogenenated catalytically according to b). The residue was triturated with ether and dried under highly reduced pressure. 5.45 g of amorphous H-Leu-Arg-Pro-cyclopropyl amide · 2 HCl were obtained that were contaminated by salts. (4.96 g = 100 %, calculated on H-Arg-Pro-cyclopropyl amide · 2 HCl). The total of the substance was dissolved together with 1.35 g of HOBt, 2.6 ml of N-ethyl morpholine and 4.4 g of Z-D-Leu-OTcp in 20 ml of dimethyl formamide. The solution was allowed to stand for 2 hours, it was concentrated and the residue was triturated twice with saturated $NaHCO_3$-solution, it was dissolved in methylene chloride, dried over Na_2SO_4, concentrated and the residue was triturated twice with saturated $NaHCO_3$-solution, it was dissolved in methylene chloride, dried over Na_2SO_4, concentrated and the residue was triturated with ether. Yield: 4.15 g (62 %, calculated on H-Arg-Pro-cyclopropylamide · 2 HCl). The substance was amorphous and was further worked without purification.

f) Z-Ser-Tyr(Bzl)-OH

To a suspension of 5.52 g (20 mmoles) of H-Tyr(Bzl)-OH in 60 ml of dimethyl formamide were added 7.68 g of Z-Ser-OBt and the solution was stirred for 6 hours at room temperature. The undissolved substances were suction-filtered and to the filtrate cooled to 0°C 300 ml of water were added. The precipitate was suction-filtered, washed with dimethylacetamide/water mixture (1:10) and water and stirred with 1 N H_2SO_4. The mixture was suction-filtered again and washed with water and dried. Recrystallization followed from ethyl acetate/petroleum ether. Yield: 7.35 g (75 %), melting point: 166°C, $[\alpha]_D^{20}$ = +20.9° (c=1, in methanol).

g) H-Ser-Tyr-D-Leu-Leu-Arg-Pro-cyclopropyl amide · 2 HCl

3.35 g (5 mmoles) of Z-D-Leu-Leu-Arg-Pro-cyclopropyl amide were dissolved in methanol and hydrogenated catalytically according to b). The residue was triturated with ether and dried under highly reduced pressure.

Yield: 2.9 g of amorphous material (95 %)

The above 2.9 g (4.7 mmoles) of H-D-Leu-Leu-Arg-Pro-cyclopropylamide · 2 HCl were suspensed together with 2.32 g (4.7 mmoles) of Z-Ser-Tyr(Bzl)-[illegible], 635 g of HOBt and 1.22 ml of N-ethylmorpholine in 10 ml of dimethyl formamide. At 0°C, 1.04 g of DCC were added and the mixture was stirred for 1 hour at 0°C and it was allowed to stand overnight at room temperature. The precipitate was suction filtered the next day and the filtrate was concentrated. The residue was triturated twice with saturated $NaHCO_3$-solution, dissolved in methylene chloride and the solution was dried over Na_2SO_4 and concentrated. The residue was triturated with ether and suction-filtered. 3.5 g (71 %) of amorphous

Z-Ser-Tyr(Bzl)-D-Leu-Leu-Arg-Pro-cyclopropyl amide were obtained which were hydrogenated catalytically according to b). The residue was triturated with ether and dried under highly reduced pressure. Yield: 292 g (= 70 %, calculated on Z-Ser-Tyr(Bzl)-OH). The crude substance was purified by distribution chromatography on the column described below:

Contents of the column: 400 ml of glacial acetic acid, 800 ml of n-butanol and 4 liters of water were shaken. 300 ml of the upper phase were stirred with 240 g of Sephadex LH 20(R). The total amount of solvent was absorbed. The column filling so pretreated was suspensed in a corresponding amount of the lower phase. The mixture was allowed to swell for 3 hours and the column 1 m x 4 cm was filled. The lower phase was used for elution.

Yield: 1.3 g of chromatographically pure substance.

$[\alpha]_D^{22}$ = -43.8° (c=1, in methanol)

h) Glu-His-Trp-Ser-Tyr-D-Leu-Leu-Arg-Pro-cyclopropyl amide

To a solution of 500 mg of Glu-His-Trp-NH-NH_2 in 6 ml of dimethyl formamide were added at -30°C 0.66 ml of a 6.05 N HCl/dioxane solution and 1.2 ml of a 10 % tert.butylnitrile solution in absolute dioxane. The solution was stirred for 20 minutes at -10°C and at -40°C, 860 mg of H-Ser-Tyr-D-Leu-Leu-Arg-Pro-cyclopropyl amide. 2 HCl and 0.78 ml of N-ethyl morpholine were added. The mixture was allowed to stand overnight at 4°C in the refrigerator, it was concentrated and the residue was triturated with ether. The substance was dissolved in water and chromatographed over Dowex(R) 1 x 2 (acetate form). The eluate was concentrated and purified over a carboxymethyl cellulose column (90 x 1.5 cm) that was equilibrated with 0.002 m of NH_4-acetate solution. The substance was added as a solution to a 0.002 m NH_4-acetate

solution. Elution was effected with a 0.002 m NH_4-acetate solution in which a gradient of a 0.1 m NH_4-acetate solution was established (mixod volume: 250 ml).

The fractions that contained the desired peptide were lyophilised twice. Yield: 405 mg of chromatographically pure substance. The content of peptide base was 77 % as per UV-spectrum (yield: 26 %). $[\alpha]_D^{20} = -39.9°$ (c=1, in dimethyl acetamide).

E X A M P L E 2

Glu-His-Trp-Ser-Tyr-D-Leu-Ser(Bu^t)-Arg-Pro-cyclopropyl amide

a) H-Ser(Bu^t)-Arg-Pro-cyclopropyl amide · 2 HCl

To a solution of 2.3 g (6 mmoles) of H-Arg-Pro-cyclopropyl amide · 2 HCl and 810 mg of HOBt in 10 ml of dimethyl formamide were added 1.56 ml of N-ethylmorpholine and 3.12 g of Z-Ser(Bu^t)-OTcp and the solution was stirred for 2 hours at room temperature. It was concentrated in vacuo, the residue was dissolved in ethyl acetate and the solution was shaken twice with saturated $NaHCO_3$-solution, dried over Na_2SO_4 and concentrated. The residue was triturated with ether and dried under highly reduced pressure. 2.4 g of amorphous substance were obtained that were hydrogenated catalytically in methanol according to Example 1 b). Yield: 2.8 g (88 %) of amorphous substance that was not uniform according to thin-layer chromatography (contaminated by about 3 by-products).

b) H-D-Leu-Ser(Bu^t)-Arg-Pro-cyclopropyl amide · 2 HCl

To a solution of 2.62 g (5 mmoles) of H-Ser(Bu^t)-Arg-Pro-cyclopropyl amide · 2 HCl and 675 mg of HOBt in 5 ml of dimethyl formamide were added at 0°C 1.3 ml of N-ethyl morpholine and 2.22 g of Z-D-Leu-OTcp. The mixture was allowed to stand overnight, it was concentrated and the residue was dissolved in ethyl acetate.

The solution was shaken twice in saturated $NaHCO_3$-solution, dried over Na_2SO_4 and concentrated. The residue was triturated with ether and dried under highly reduced pressure. Yield: 2.4 g of amorphous substance that was hydrogenated catalytically in methanol according to Example 1 b). The residue was triturated with ether. Yield: 2.1 g of an amorphous substance (66 %, calculated on Z-D-Leu-OTcp. The substance was further worked without purification.

c) H-Trp-Ser-Tyr-D-Leu-Ser(Bu^t)-Arg-Pro-cyclopropylamide · 2 HCl

To a solution of 1.99 g (3.3 mmoles) of Z-Trp-Ser-Tyr-NH-NH_2 in 25 ml of dimethylformamide were added at -30°C, 2.18 ml of a 6.05 N HCl/dioxane-solution and 3.97 ml of a 10 % tert.-butylnitrite solution in absolute dioxane. The mixture was stirred for 20 minutes at -10°C and at -40°C, 2.6 ml of N-ethyl morpholine and 2.1 g (3.3 mmoles) of H-D-Leu-Ser(Bu^t)-Arg-Pro-cyclopropylamide · 2 HCl were added. The mixture was allowed to stand overnight at 4°C in the cooling chamber, it was concentrated and the residue was triturated with ether. It was hydrogenated according to Example 1 b) and the crude substance was purified chromatographically on Sephadex LH 20 according to Example 1 g). Yield: 1.25 g (35 %) of chromatographically pure amorphous substance.

d) [Glu-His-Trp-Ser-Tyr-D-Leu-Ser(Bu^t)-Arg-Pro-cyclopropylamide-diacetate

To a solution of 538 mg (0.5 mmole) of H-Trp-Ser-Tyr-D-Leu-Ser(Bu^t)-Arg-Pro-cyclopropylamide · 2 HCl, 151 mg (0.5 mmole) of [Glu-His-OH and 135 mg of HOBt in 5 ml of dimethyl formamide were added at 0°C, 0.13 ml of N-ethyl morpholine and 110 mg of DCC. The mixture was allowed to stand for 1 hour at 0°C and overnight

at room temperature, the precipitate was suction-filtered, concentrated and the residue was triturated with ether. It was dissolved in water, undissolved substances were filtered off and purification followed according to Example 1 h) over Dowex 1 x 2 and carboxymethyl cellulose and, subsequently, over Sephadex LH 20 according to Example 1 g). Yield: 165 mg. The content of peptide base was 78 % as per UV-spectrum (yield of 20.5 %).

$[\alpha]_D^{23}$ = -28.9° (c=1, in dimethyl acetamide)

E X A M P L E 3 (Preparation for oral adminstration)

10 g of pGlu-His-Trp-Ser-Tyr-D-Leu-Leu-Arg-Pro-cyclopropyl amide diacetate were triturated with 542 g of lactose. The trituration was mixed with 300 g of potato starch, it was moistured with the alcoholic solution of 8 g of gelatin and granulated. After drying, 60 g of potato starch, 10 g of magnesium stearate, 20 g of highly disperse silicium oxide and 60 g of talcum were added and the mixture was compressed to 10,000 tablets of each 150 mg weight. Each tablet contained 1 mg of active substance.

E X A M P L E 4 (Preparation for intranasal administration)

4 g of pGlu-His-Trp-Ser-Tyr-D-Leu-Ser(Bu^t)-Arg-Pro-cyclopropylamide-diacetate were dissolved in 100 ml of distilled water. At the same time, 31.2 g of $NaPHO_4 \cdot 2\ H_2O$, 66.29 g of Na_2HPO_4, 25 g of sodium chloride and 100 g of benzyl alcohol were dissolved in 8 l of distilled water and 500 g of polyvinyl alcohol having a K-value of about 90 were added. The two solutions were combined and filtered. The single dose of 20 µg was contained in 0.05 ml.

E X A M P L E 5 (Preparation for intranasal adminstration)

100 g of anhydrous Lanolin and 440 g of Vaseline were molten together. To the cold melt a suspension of 800 mg of microfine

Glu-His-Trp-Ser-Phe-D-Leu-Ser(Bu^t)-Arg-Pro-cyclopropyl amide diacetate in 359.2 g of liquid paraffin was added. Finally, 10 g of benzyl alcohol were added and the ointment was homogenized. The single dose of 40 µg was contained in 0.05 g of ointment.

EXAMPLE 6 (Preparation for injections)

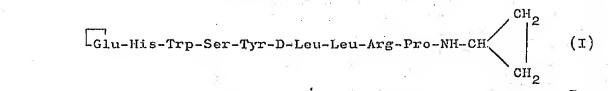
2 mg of [Glu-His-Trp-Ser-Tyr-D-Leu-Leu-Arg-Pro-cyclopropyl-amide-diacetate were dissolved in 500 ml of bidistilled water to which 100 ml of phosphate buffer of pH 4.5 were added. 1 g of mannitol and the calculated amount of NaCl was added to obtain isotonicity and water was added to a volume of 1 liter. After filtration under sterile conditions, ampoules of 1 ml or 2 ml were filled and lyophilized.

EXAMPLE 7 (Preparation for injections)

The example was carried out according to Example 6, however, before filling with water, 2.5 g of 4-hydroxybenzoic acid methyl ester were added. After filtration under sterile conditions, ampoules of 1 ml or 2 ml were filled.

What is claimed is: DEFINING THE INVENTION ARE /

1) Peptides of the formula I



in which, optionally Tyr can be replaced by Phe and Leu^7 by $Ser(Bu^t)$, $Asp(OBu^t)$, $Glu(OBu^t)$, $Cyr(Bu^t)$, Lys(Boc) or Orn (Boc).

2) Process for the preparation of peptides as claimed in claim 1, which comprises preparing them

a) by fragment condensation of peptide fragments usual in peptide chemistry according to the condensation scheme

1 - 3 + 4 - 10 or 1 - 2 + 3 - 10 or

b) by stepwise synthesis,

in which case other functional groups are blocked intermediately by protective groups capable of being split off by hydrogenation, or protective groups capable of being split off in an alkaline or slightly acid medium.

3) Pharmaceutical preparations for peroral, intranasal, intramuscular, subcutaneous or intravenous administration consisting of or containing compounds as claimed in claim 1.

4) Pharmaceutical preparations for peroral administration consisting of or containing compounds as claimed in claim 1.

5) Process for the preparation of pharmaceutical compositions as claimed in claim 3, in which compounds as claimed in claim 1 are brought, optionally together with pharmaceutically suitable carriers and/or stabilizers into a therapeutically suitable administration form.

Dated this 6 day of August 1975

HOECHST AKTIENGESELLSCHAFT

[illegible] WATERS & SONS
PATENT ATTORNEYS
50 QUEEN STREET
MELBOURNE
AUSTRALIA